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# Diversity of *Fusarium* Species Associated with Post-harvest Fruit Rot Disease of Tomato

(Kepelbagaian Spesies *Fusarium* yang Berasosiasi dengan Penyakit Lepas Tuai Reput Buah Tomato)

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ABSTRACT

Fusarium *species is one of the common pathogens of post-harvest disease to cause rot on tomato and other perishable vegetable fruits. The objectives of this study were to determine the diversity of* Fusarium *isolated species from post-harvest diseases of tomato fruit, to identify the causal organisms by using phenotype characteristics and to verify the pathogens of* Fusarium *fruit of tomato based on pathogenicity test. Carnation leaf-piece agar (CLA) and potato dextrose agar (PDA) media were used for phenotype-based identification of the* Fusarium *isolates with emphasis for characterizations of the shapes and sizes of the macroconidia and microconidia, colony features, growth rates, conidiogenous cells and chlamydospores. A total of 180* Fusarium *isolates were obtained from 13 locations throughout Selangor.* Fusarium solani *was most abundantly isolated (34%) followed by* F. semitectum *(31%) and* F. oxysporum *(31%),* F. subglutinans *(3%) while the least was* F. equiseti *(1%). Twenty seven isolates were tested for pathogenicity test by injecting 1 mL of the conidial suspension onto healthy tomatoes. All the tested* Fusarium *isolates were pathogenic on tomato with different severity levels. The non-inoculated controls showed no symptoms of fruit rot. The most virulent was* F. oxysporum *isolate B711T with DSI 93.75%, while the least were isolates of* F. solani *(B647T) and* F. oxysporum *(B727T) with DSI 37.5%. Majority of the isolated* Fusarium *species can potentially produce mycotoxins as their secondary metabolites. The potential production of mycotoxins by pathogenic isolates of* Fusarium *species in contaminated tomato fruits could pose health hazards when consumed.*

*Keywords: Disease severity index; fruit;* Fusarium; *pathogen; post-harvest; tomato*

ABSTRAK

*Spesies* Fusarium *merupakan salah satu patogen biasa lepas tuai yang menyebabkan penyakit reput buah tomato, sayuran dan buah lembut yang lain. Objektif kajian ini adalah untuk mengenal pasti kepelbagaian spesies* Fusarium *yang dipencilkan daripada penyakit reput lepas tuai tomato, mengenal pasti organisma penyebab berasaskan ciri fenotip dan mengesahkan patogen penyakit reput* Fusarium *tomato berasaskan ujian kepatogenan. Agar keratan daun teluki (CLA) dan agar kentang dekstrosa (PDA) digunakan untuk pengenal pastian isolat* Fusarium *berasaskan ciri fenotip yang menumpukan bentuk dan saiz makrokoni diadanmikrokonidia, ciri koloni, kadar pertumbuhan, sel konidiogen dan klamidospora. Sejumlah 180 isolat* Fusarium *diperoleh dari 13 lokasi sekitar Selangor.* F. solani *adalah yang terbanyak dipencilkan (34%) diikuti oleh* F. semitectum *(31%),* F. oxysporum *(31%),* F. subglutinans *(3%), di mana yang paling sedikit adalah* F. equiseti *(1%). Dua puluh tujuh isolat* Fusarium *diuji dalam ujian kepatogenan dengan menyuntik 1 mL ampaian konidia ke atas tomato sihat. Kesemua isolat* Fusarium *yang diuji adalah patogenik ke atas tomato pada tahap keseriusan yang berbeza. Tomato yang tidak diinokulat tidak menunjukkan sebarang simptom reput buah. Isolat B711T* F. oxysporum *merupakan isolat yang paling virulen dengan DSI 93.75%, sementara yang paling lemah adalah isolat* F. solani *(B647T) dan* F. oxysporum *(B727T) dengan DSI 37.5%. Majoritis pesies* Fusarium *yang dipencilkan berpotensi menghasilkan mikotoksin sebagai metabolit sekunder. Kebolehan menghasilkan mikotoksin oleh isolat yang patogenik pada buah tomato yang dijangkiti boleh menyebabkan keracunan dan masalah kesihatan jika dimakan.*

*Kata kunci:* Fusarium*; indeks keseriusan penyakit; lepas tuai; patogen; penyakit reput buah; tomato*

## Introduction

Vegetable fruits are highly perishable crops. Thus, post-harvest handling, storage, transportation and marketing are seriously affecting their quality in the market, where improperly handling, packaging, storage and transportation could cause decay and increase the production of micro-organisms due to changing physiological state of the fruits and vegetables (Wilson et al. 1991). Fruits, due to their low pH, high moisture content and nutrient composition are very susceptible to attack by pathogenic fungi, which in addition to causing rots may also make them unfit for consumption by producing mycotoxins (Moss 2002).

Tomato fruit rots are mainly caused by bacteria and fungi. However, some plant pathogens, viruses and nematodes could also be responsible for post-harvest losses but do not cause progressive deterioration of tomatoes in storage. Several fungal species can cause fruit decay in tomatoes such as the sour rot pathogens caused by *Geotrichum candidum*, Rhizopus rot caused by *Rhizopus stolonifer*, buckeye rot caused by *Phytophthora* species, black mold rot caused by *Alternaria arborescens* and Fusarium rots caused by *Fusarium* species (Mahovic et al. 2004).

*Fusarium* species can appear as saprophytes or pathogens on plants, animals as well as humans. They are well associated with a wide range of plants in their natural habitats such as tomato, banana, asparagus, barley, mango, pineapple, carnation, coffee, corn, grasses, legumes, oats, pine, rice, sorghum, sugarcane and wheat (Burgess et al. 1994; Nelson et al. 1990). Harvested tomatoes are susceptible towards infections caused by *Fusarium* species due to its succulent epicarp which enable the fungal hyphae to penetrate deeply into the fruit (Tournas & Katsoudas 2005). As a result, the yield of this economically important farm product is affected, hence lowering the production rate (Salleh & Mushitah 1991). *Fusarium* rot on tomato fruits are often caused by *Fusarium* species (Denis 1983; Mehrotra 1989; Sherf & Macnab 1986; Solunkhe & Desai 1984) and the disease symptoms include rots softer and extend into the center of the fruit. The rotted tissue is often water-soaked and becomes covered by white, yellow or pinkish mycelium externally while the infected tissue is discolored and appears pale brown (Denis 1983). The disease causes the vegetable fruits unmarketable as consumer will only choose those that are fresh and healthy (Nurulhuda et al. 2009). Hence, *Fusarium* species have been isolated from decaying tomato fruits (Oladiran & Iwu 1992). Tomato contaminated with *Fusarium* species are lethal for human and animals consumption since several species of them produce mycotoxins (Burgess 1985; Joffee 1986; Nelson et al. 1990).

In Malaysia, many *Fusarium* species had been isolated from various vegetable fruits but are not well documented with detailed information on its diversity on tomato. Therefore, the objectives of this study were to determine the diversity of *Fusarium* species from *Fusarium* fruit rot (FFR) of tomato, to tentatively identify the causal organisms based on phenotype characteristics and to verify the pathogens of the disease by pathogenicity test.

## Materials and Methods

### Sampling and Fungal Isolation

Samples of tomato with post-harvest fusarium fruit rot symptoms were collected from 13 locations throughout Selangor. All samples were surface sterilized in 10% chlorox® and washed in sterile distilled water for 2 min each. The washed tomatoes were bolt dried using filter paper and transferred onto peptone pentachloronitrobenzene agar (PPA) media (Papavizas 1967). The samples were subsequently incubated at room temperature for 7 days. Single spore isolation was carried out onto potato dextrose agar (PDA) by streaking technique. Single spored colony was transferred onto new PDA to obtain pure culture and incubated at room temperature for 7 days.

### Identification of *Fusarium* Species

Pure cultures of the isolates were cultured onto carnation leaf-piece agar (CLA) for phenotype-based identification following *Fusarium* manual by Leslie and Summerell (2005). Cultures on CLA were incubated at standard condition which is room temperature (25±2°C) for 7 days. The isolates phenotype examined for identification were shapes and sizes of macroconidia and microconidia; number of septa and shapes of the apical and basal cells of the macroconidia, conidiogenous cells, growth rate, presence of chlamydospore, colony colour, growth and pigmentation on PDA.

### Diversity Index of the *Fusarium* Species

The Shannon-Weiner index for diversity of the *Fusarium* species in each location was calculated using the formula by Venco and Meneses (1996):

$$H' = \sum_{i=1}^{S} pi(\ln pi),$$

where H' is the value of Shannon-Weiner Index, S is the number of species in the community, Pi is the is the relative abundance of the species in the community and ln is the natural log.

### Virulence Test

Virulence test was conducted to verify the *Fusarium* isolates as pathogens or saprophytes on tomato fruits. Mature, fresh and healthy tomato fruits were surface sterilized and washed in the sterile distilled water both for 2 min each and subsequently placed on filter paper for drying. Four replicates for each of 27 selected isolates were used and were resub-cultured onto PDA and incubated at room temperature (25±2°C) for 7 days. The conidia were harvested using 20 mL sterile distilled water and only 1 mL conidial suspension was injected into the inner tissue of the tomato using the sterile syringe. The inoculated fruits were incubated for 7 days along with the uninoculated fruits as controls. The test was repeated 3 times. All inoculated and uninoculated tomatoes were kept in an individual sterilized container, 2 pieces/container with a cover and incubated at room temperature condition. The fungal colony from disease lesion was subcultured on PPA and PDA. The isolated fungal were identified based on their morphological characteristics including colony features, growth rates and pigmentation. Those isolated fungal that appeared the same as the inoculum were confirmed their pathogenicity on tomato fruits.

### DISEASE ASSESSMENT

The disease symptoms were scored based on a disease scale from 0 to 4 devised by Amadi et al. (2009) with a slight modification for tomato. All scales were characterized with a particular symptom on tomato fruit rot labelled on it as shown in Table 1. The DSI data was then analysed using Friedman test of the non-parametric test of SPSS programme at $p<0.05$ in order to compare the variation of the DSI distribution among the isolates.

## RESULTS AND DISCUSSION

A total of 180 isolates of *Fusarium* were isolated from post-harvest disease of tomato fruit rot. Identification of all the isolates were based on phenotypic characterization. Five *Fusarium* species were identified namely, *F. solani, F. semitectum, F. oxysporum, F. subglutinans* and *F. equiseti*. Table 2 shows the phenotype of the five *Fusarium* species isolated from tomato throughout Selangor. Figure 1 shows the microscopic characteristics of the *F. solani, F. semitectum, F. oxysporum, F. subglutinans* and *F. equiseti*.

Table 3 shows the distribution of *Fusarium* isolates obtained from post-harvest *Fusarium* fruit rot of tomatoes in Selangor and their diversity based on Shanon-Weiner index. The highest distribution was in Sri Serdang with 77 *Fusarium* isolates followed by Kajang (39 isolates) while 8 *Fusarium* isolates each for Semenyih, Balakong, Shah Alam, Seri Kembangan and Ampang. The fewest was scored in Selayang, Puchong, Bandar Sunway, Subang Jaya, Rawang and Sungai Buloh with only 4 isolates. *F. solani* (34%) was dominantly isolated followed by *F. semitectum* (31%), *F. oxysporum* (31%), *F. subglutinans* (3%) and *F. equiseti* (1%). The community diversity of the *Fusarium* species in each location was however computed using Shannon-Weiner Index(H') with the highest value of the Shannon-Weiner index was recorded in Sri Serdang (H'= 0.363) followed by Kajang (H' = 0.332). The value of (H' = 0.137) each for Semenyih, Balakong, Shah Alam, Seri Kembangan and Ampang while the least value recorded in Selayang, Puchong, Bandar Sunway, Subang Jaya, Rawang and Sungai Buloh (H'= 0.084).

*F. solani* was the most common recovered *Fusarium* from tomato fruit rot. In Malaysia, *F. solani* has been reported as a plant pathogen infecting tomato and other vegetable fruits (Nurulhuda et al. 2009) and was isolated from soil and also grasses where 39% isolated from cultivated and non-cultivated soil (Nik Mohd Izham 2008), 21.3% isolated from soil cultivated with cucurbits (Siti Nordahliawate et al. 2009) and 10.3% isolated from grasses (Nur Ain Izzati et al. 2009). In addition, *F. solani* was also reported to have been recovered from corn (Nur Ain Izzati et al. 2011) and root and stem rot of *Dendrobium* orchids (Latiffah et al. 2009). Besides damaging tomato fruits in storage (Amadioha & Uchendu 2003), *F. solani* was identified as a pathogen of a number of tropical agricultural crops such as potato, soybeans, peas and peppers (Grunwald et al. 2003). Leslie and Summerell (2005) reported that *F. solani* produces mycotoxins that are lethal to humans and animals health. *F. solani* has been known to synthesize compounds such as fusalanipyrone (Abraham et al. 1990), fusaric acid (Bacon et al. 1996), moniliformin (Chelkowski et al. 1990) and moreover, a number of chemically unidentified toxic compounds are produced by *F. solani* but its ability to produce trichothecene and zearalenone have not been confirmed (Marasas et al. 1984).

*F. semitectum* and *F. oxysporum* were the second most isolated *Fusarium* from decaying tomato fruits. *F. semitectum* was reported to have been isolated from decaying okra, bitter gourd, loofah, red chilli and cucumber (Nurulhuda et al. 2009), corn (Nur Ain Izzati et al. 2011) and dragon fruit (*Hylocereus polyrhizus*) (Hawa et al. 2010). *F. semitectum* was however reported to have caused diseases on banana fruits (Wallbridge 1981), melons (McGovern 1994), beans (Dhingra & Muchovej 1979), sorghum (Gopinath et al. 1985), walnut (Seta et al. 2004) and storage rot of mushrooms (Seth & Shandilya 1978). *F. semitectum* has also been known to produce mycotoxins such as trichothecene and zearalenone (Marasas et al. 1984), beauvericin (Logrieco et al. 1998) and moniliformin (Rabie et al. 1982).

*F. oxysporum* was reported to have been isolated from tomato fruits (Fajola 2004), okra, bitter gourd and tomato (Nurulhuda et al. 2009), corn (Nur Ain Izzati et al. 2011) and *Eurycoma longifolia* Jack (Nur Ain Izzati & Siti Nordahliawate 2010). Besides, it was also isolated from decay area of tomato fruit and the decay area appeared to

TABLE 1. Disease severity index (DSI) for disease assessment with a slight modification for tomato (Amadi et al. 2009)

| Disease scale | Description | Inference |
|---|---|---|
| 0 | No visible symptoms on fruits | No infection |
| 1 | 1 – 25% of inoculated area covered with slight necrotic | Mild infection |
| 2 | 26 – 50% of inoculated area covered with necrotic and white mycelia of fungal | Moderate infection |
| 3 | 51 – 75% of sample are necrotic with spore mass appeared | Severe infection |
| 4 | >76% necrotic tissue appears soft and decay with fungal mass | Very severe/Devastating |

$$DSI = \frac{\sum(A \times n)}{\sum B\ 5} \times 100$$

A = disease scale (0, 1, 2, 3 or 4); N = number of tomato for each disease scale and B = total number of tomatoes

TABLE 2. Morphological characteristics of *Fusarium* species isolated from post-harvest tomato fruits

| *Fusarium* species | Characteristics | | | | | |
|---|---|---|---|---|---|---|
| | Macroconidia | Microconidia | Conidiophores | Chlamydospore | Pigmentation on PDA | Growth rate at 27°C |
| *F. solani* | 5-septate, wide, straight, robust and stout. Apical cell is blunt and rounded foot shaped basal cell | Oval and reniform shaped with 0 – 1 septa and abundant in the aerial mycelia | Long monophialide | Globose and exist singly with smooth and rough walled | White tinged with brown | 1.7-5.4 cm |
| *F. semitectum* | 5-septate and thin-walled with a curved dorsal surface and a slightly straighter ventral surface. Tapered apical cell and foot shaped basal cell | Oval and kidney shaped with 0-1 septa and scarce in the aerial mycelia | Monophialide and polyphialide | Globose, smooth and formed singly and in chains intercalary and terminal | Brown | 2.1-5.8 cm |
| *F. oxysporum* | 5 septate, wide and thin-walled with a curved dorsal surface and a straighter ventral surface. Apical cell is tapered and foot shaped basal cell | Oval and kidney shaped with 0-1 septa and usually abundance in false heads mycelia | Short monophialide | Oval and occurred singly and in chains with smooth and rough walled intercalary and terminal | White tinged with purple | 3.1-5.6 cm |
| *F. subglutinans* | 3 septate, wide and thin-walled with dorsoventral surfaces slightly parallel. Curved apical cell and foot shaped basal cell | Oval, single-celled, abundant and also occurred in false heads only | Monophialide and polyphialide | Absent | Dark purple | 2.1-6.1 cm ) |
| *F. equiseti* | 5 septate, long, thick-walled and sickle-shaped. Tapered and whip-like apical cell and elongated foot shaped basal cell | Scanty and produce in false heads | Monophialide | Oval shaped and occurred singly, in chains and in clumps intercalary and terminally | Dark brown | 2.7-5.5 cm |

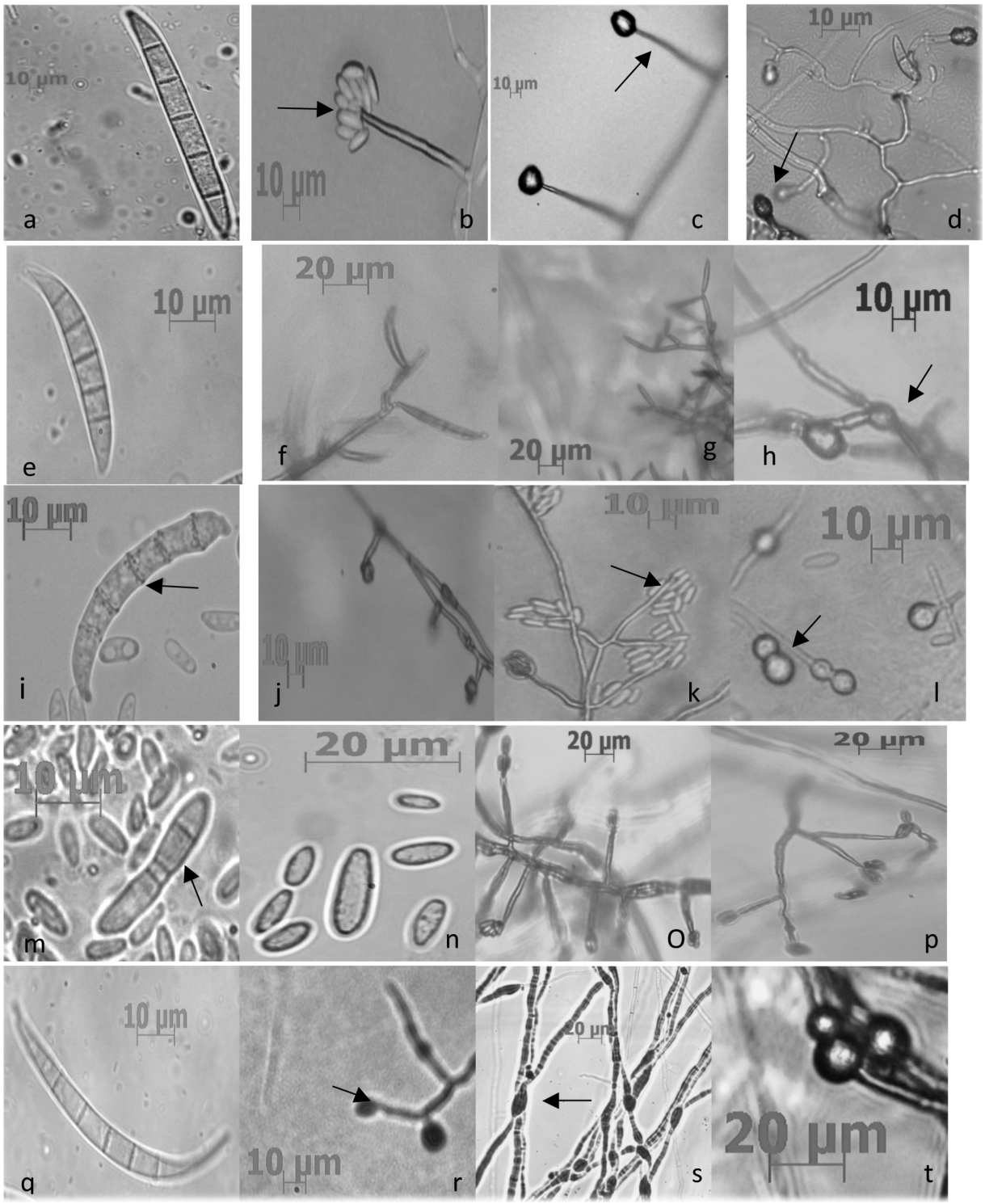



FIGURE 1. Microscopic characteristics of *Fusarium* species isolated from post-harvest disease of tomato *F. solani* a. (Macroconidia) b. (Microconidia in false heads) c. (Monophialide) & d. (Chlamydospore); *F. semitectum* e. (Macroconidia) f. (Monophialide) g. (Polyphialide) & h. (Chlamydospores in chain); *F. oxysporum* i. (Macroconidia) j. (Monophialide) k. (Microconidia in false heads) & l. (Chlamydospores in chains); *F. subglutinans* m. (Macroconidia) n. (Microconidia) o. (Monophialide) & p. (Polyphialide); *F. equiseti* q. (Macroconidia) r - s. (Microconidia in false heads) & t (Clustered chlamydospores)

TABLE 3. Distribution of *Fusarium* species isolated from post-harvest disease of tomato in Selangor and their diversity based on Shanon-Weiner index

| Location | Number of isolate | | | | | Total | H' |
|---|---|---|---|---|---|---|---|
| | *F. solani* | *F. semitectum* | *F. oxysporum* | *F. subglutinans* | *F. equiseti* | | |
| Sri Serdang | 28 | 31 | 16 | 1 | 1 | 77 | 0.366 |
| Selayang | 0 | 0 | 0 | 4 | 0 | 4 | 0.084 |
| Kajang | 10 | 5 | 24 | 0 | 0 | 39 | 0.332 |
| Semenyih | 4 | 0 | 4 | 0 | 0 | 8 | 0.138 |
| Puchong | 0 | 0 | 4 | 0 | 0 | 4 | 0.084 |
| Balakong | 4 | 4 | 0 | 0 | 0 | 8 | 0.138 |
| Shah Alam | 0 | 8 | 0 | 0 | 0 | 8 | 0.138 |
| Bandar Sunway | 4 | 0 | 0 | 0 | 0 | 4 | 0.084 |
| Subang Jaya | 4 | 0 | 0 | 0 | 0 | 4 | 0.084 |
| Seri Kembangan | 0 | 0 | 8 | 0 | 0 | 8 | 0.138 |
| Ampang | 0 | 8 | 0 | 0 | 0 | 8 | 0.138 |
| Rawang | 4 | 0 | 0 | 0 | 0 | 4 | 0.084 |
| Sungai Buloh | 4 | 0 | 0 | 0 | 0 | 4 | 0.084 |
| Total | 62 | 56 | 56 | 5 | 1 | 180 | 1.889 |
| Percentage (%) | 34.44 | 31.11 | 31.11 | 2.78 | 0.56 | 100 | |

be water-soaked and slightly sunken (Snowdon 1991). *F. oxysporum* moreover causes decay on tomato fruits (Yeni et al. 2010), wilting, crown and root rot on tomato plants and symptoms of such infections are stunted growth, older leaves are often curved downward and subsequently the plants wilt and die (Denis et al. 1994; Momol et al. 2008). While crown and root rot stunted growth, yellowing and premature loss of cotyledons and lower true leaves, brown lesion, root rot, wilting and finally the death of the plants (Momol et al. 2008). *F. oxysporum* has also been reported to produce zearalenone and trichothecene (Marasas et al. 1984), beauvericin (Moretti et al. 2002) and moniliformin (Abbas et al. 1989).

*F. subglutinans* was the fourth *Fusarium* species isolated from tomato fruit rot in this study. It has been isolated from okra and red chilli (Nurulhuda et al. 2009), peppers (Mathur & Utkhede 2004), banana (Jimenez et al. 1993), rice (Nur Ain Izzati & Salleh 2009), sugar cane (Siti Nordahliawate et al. 2008), corn (Nur Ain Izzati et al. 2011), soybean (Schlub et al. 1981), millet (Onyike et al. 1991) and sorghum (Saubois et al. 1999). *F. subglutinans* produces beauvericin (Leslie et al. 2004), moniliformin (Lew et al. 1996) and little or no fumonisins (Reynoso et al. 2004).

*F. equiseti* was the least *Fusarium* isolated from post-harvest fruit rot of tomato and has been reported as a plant pathogen infecting tomato (Fajola 2004; Oladiran & Iwu 1992), pumpkin (Elmer 1996) and cucurbit (Adams et al. 1987) and moreover recovered from okra and cucumber (Nurulhuda et al. 2009) and corn (Nur Ain Izzati et al. 2011). *F. equiseti* was also reported to produce zearalenone (Hestbjerg et al. 2002), beauvericin (Logrieco et al. 1998) and trichothecene (Marasas et al. 1984).

In the virulent test, a total of 108 tomato fruits were inoculated with 27 isolates of the *Fusarium* (Table 4) with the control fruits. All the inoculated and non-inoculated fruits were observed from day after inoculation (dai) 1 to 7 every 24 h. On dai 1, there was no growth either of necrosis or mycelia on the epicarp of the majority of tomato fruits (0% DSI) except in 4 isolates with 6 - 7% DSI as shown in Table 5. On dai 2 and 3, there were growth and development of necrosis on most of the fruits (22 isolates with 7 - 25% DSI) with few exception associated with white mycelia of fungal (5 isolates with 37 - 50% DSI). The fungi were penetrated into the epicarp and lied within the fleshy parts of the mesocarp. The necrosis of the epicarp enlarges outwards and the fruit becomes water-soaked as the infestation of the fungi continued as revealed in Figure 2(a) and 2(b). Moderate infection dominated majority of the inoculated tomato fruits (18 isolates with 40 - 60% DSI) and others in few instances with spore mass appeared (5 isolates with 60 - 75% DSI) on dai 4 and 5. Isolates recorded with lower DSI were B645T (25%) B647T (37.5%), B721T (37.5%) and B727T (31.25%). Severe infection covered almost half of the fruits (12 isolates with 60 - 75% DSI) and few others very devastating in which their necrotic tissues were soft and decay with fungal mass appeared on dai 6 and 7 (11 isolates with >80% DSI). Isolates recorded with very lower DSI were B645T (43.75%), B647T (37.5%), B689T (56.25%) and B727T (37.5%). In severe devastating, the fungi extended deep into the seed cavity to destroying the seed coat and cotyledons, Figure 2(c). The non-inoculated controls showed no symptoms of fruit rot from dai 1 to 7.

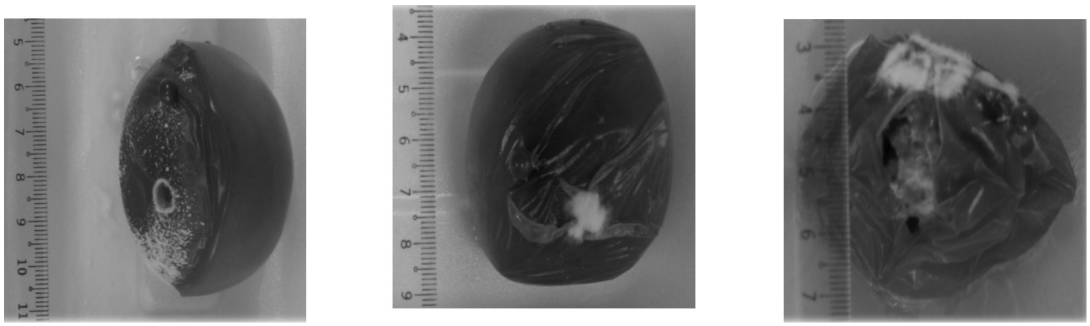
The DSI data obtained from dai 1 - 7 was then analysed using Friedman test of the Non-parametric test of SPSS programme. The results revealed that there are no difference in the distributiouns of isolate, DSI and day. The DSI distributions are significantly different among the isolates ($p<0.05$). The DSI data for dai 7 depicted that the *F. oxysporum* was the most devastating on tomato fruit rot (B711T - 93.75%) followed by *F. solani* (B684T - 87.50%), *F. subglutinans* (87.50%), *F. semitectum* (4 isolates with 75%) and *F. equiseti* (B607T - 68.75%) while the least severe however were recorded on *F. solani* (B647T - 37.50%) and *F. oxysporum* (B727T - 37.50%) (Table 5).

TABLE 4. Source of isolates used in the pathogenicity test

| *Fusarium* species | Isolate identity | Locality |
|---|---|---|
| *F. semitectum* (5 isolates) | B602T | Sri Serdang |
| | B605T | Sri Serdang |
| | B609T | Sri Serdang |
| | B649T | Sri Serdang |
| | B675T | Sri Serdang |
| *F. solani* (8 isolates) | B616T | Sri Serdang |
| | B627T | Sri Serdang |
| | B647T | Sri Serdang |
| | B664T | Sri Serdang |
| | B684T | Kajang |
| | B698T | Kajang |
| | B721T | Semenyih |
| | B722T | Semenyih |
| *F. oxysporum* (10 isolates) | B633T | Sri Serdang |
| | B645T | Sri Serdang |
| | B688T | Kajang |
| | B689T | Kajang |
| | B711T | Kajang |
| | B713T | Kajang |
| | B717T | Semenyih |
| | B718T | Semenyih |
| | B725T | Puchong |
| | B727T | Puchong |
| *F. subglutinans* (3 isolates) | B658T | Sri Serdang |
| | B678T | Semenyih |
| | B679T | Semenyih |
| *F. equiseti* (1 isolate) | B607T | Sri Serdang |

TABLE 5. Disease severity index (DSI) for day 1 - 7

| *Fusarium* species | Isolate identity | Disease severity index (DSI %) for each day | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
| *F. semitectum* | B602T | 0 | 25.00 | 25.00 | 43.75 | 50.00 | 68.75 | 75.00 |
| | B605T | 0 | 25.00 | 25.00 | 43.75 | 50.00 | 68.75 | 68.75 |
| | B609T | 0 | 25.00 | 25.00 | 43.75 | 50.00 | 75.00 | 75.00 |
| | B649T | 0 | 25.00 | 37.50 | 50.00 | 50.00 | 75.00 | 75.00 |
| | B675T | 0 | 18.75 | 25.00 | 50.00 | 50.00 | 75.00 | 75.00 |
| *F. solani* | B616T | 0 | 25.00 | 25.00 | 50.00 | 50.00 | 75.00 | 81.25 |
| | B627T | 6.25 | 25.00 | 25.00 | 50.00 | 50.00 | 75.00 | 75.00 |
| | B647T | 0 | 12.50 | 18.75 | 18.75 | 37.50 | 37.50 | 37.50 |
| | B664T | 0 | 25.00 | 37.50 | 50.00 | 62.50 | 75.00 | 81.25 |
| | B684T | 0 | 12.50 | 25.00 | 50.00 | 75.00 | 87.50 | 87.50 |
| | B698T | 0 | 25.00 | 25.00 | 50.00 | 50.00 | 75.00 | 75.00 |
| | B721T | 0 | 18.75 | 25.00 | 25.00 | 37.50 | 50.00 | 62.50 |
| | B722T | 0 | 12.50 | 25.00 | 31.25 | 43.75 | 62.50 | 87.50 |
| *F. oxysporum* | B633T | 0 | 18.75 | 25.00 | 37.50 | 43.75 | 62.50 | 75.00 |
| | B645T | 0 | 0 | 18.75 | 25.00 | 25.00 | 43.75 | 43.75 |
| | B688T | 0 | 25.00 | 50.00 | 62.50 | 75.00 | 87.50 | 87.50 |
| | B689T | 0 | 25.00 | 25.00 | 37.50 | 43.75 | 50.00 | 56.25 |
| | B711T | 0 | 18.75 | 25.00 | 37.50 | 56.25 | 68.75 | 93.75 |
| | B713T | 0 | 25.00 | 25.00 | 37.50 | 50.00 | 62.50 | 87.50 |
| | B717T | 0 | 18.75 | 25.00 | 37.50 | 56.25 | 68.75 | 75.00 |
| | B718T | 0 | 25.00 | 25.00 | 50.00 | 50.00 | 68.75 | 81.25 |
| | B725T | 6.25 | 25.00 | 25.00 | 31.25 | 43.75 | 56.25 | 68.75 |
| | B727T | 0 | 0 | 25.00 | 25.00 | 31.25 | 31.25 | 37.50 |
| *F. subglutinans* | B658T | 0 | 25.00 | 25.00 | 43.75 | 50.00 | 68.75 | 87.50 |
| | B678T | 6.25 | 18.75 | 37.50 | 50.00 | 62.50 | 75.00 | 81.25 |
| | B679T | 6.25 | 25.00 | 50.00 | 50.00 | 75.00 | 75.00 | 81.25 |
| *F. equiseti* | B607T | 0 | 18.75 | 25.00 | 43.75 | 43.75 | 56.25 | 68.75 |



a) B647T b) B727T c)B711T

FIGURE 2. Mild to moderate infection in (a) and (b) (B647T and B727) while very severe/devastating in (c) (B711T)

All the isolates tested were pathogenic on tomato with different severity.

Tomato fruit rots caused by *Fusarium* was due to their utilization of ascorbic acid and soluble sugar contents in the fruit for their metabolic activities which showed a reduction trend with the days of incubation but there is a slight increase of soluble sugars on the 6$^{th}$ day after inoculation (Oladiran & Iwu 1992). Similarly Teisson et al. (1979) reported that fungal isolates utilized ascorbic acid in tomato fruits for their own metabolic activities. The reduction in ascorbic acid content also occurred in pineapples affected by black heart disorder (Abdullah et al. 1985, 1986). The carbohydrate content, especially the sugars, were reduced during the infection of sweet potatoes by *Botrydioplodia theobromae* (Arinze 1974). There is a decreased in sugar content of onion affected by *Pseudomonas fluorescens* (Oguntuyo 1981). The infection of pineapple fruits with black heart disorder was reported to have lower soluble solid than the uninfected fruits (Abdullah et al. 1986).

In conclusion, the present study therefore depicted that tomato fruits which form an important integral part of the human diet in providing basic food, essential vitamins, trace elements, food flavour, acceptability and could be used in many ways either in the raw or cooked state (Oladiran & Iwu 1992) are beset with decaying problems caused by micro-organisms. Fungi and bacteria have been isolated from decaying tomato fruits (Arinze 1986). The contamination of tomato fruits by fungi could pose threats on the biodiversity as the five *Fusarium* species isolated namely, *F. solani, F. semitectum, F. oxysporum, F. subglutinans* and *F. equiseti* were reported to produce mycotoxins. Mycotoxins are potentially toxigenic chemical compounds produced by some saprophytic *Fusarium* that affect humans and animals health when they entered food chains (Joffe 1986; Nelson et al. 1990). The findings of this study on the diversity of *Fusarium* species, morphological characteristics of the causal organisms and the virulence test is hope to provide an integrated information to formulating suitable control measure against the disease.

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